7.5 μg DNA

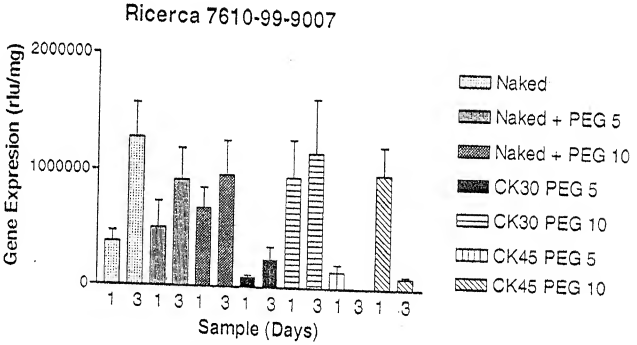


Fig. 1

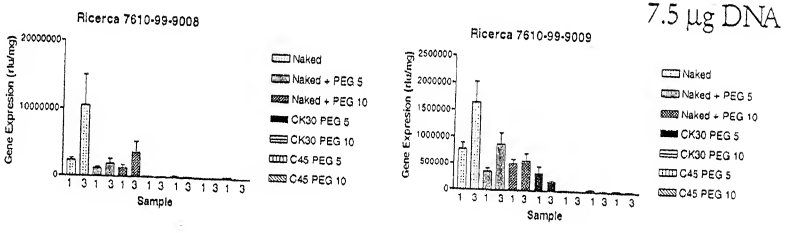

7.5 µg DNA
Ricerca 7610-99-9008
Gene Expression (rlu/mg)
20000000
10000000
0
1 3 1 3 1 3 1 3 1 3 1 3 1 3
Sample
Naked
Naked + PEG 5
Naked + PEG 10
CK30 PEG 5
CK30 PEG 10
C45 PEG 5
C45 PEG 10
Ricerca 7610-99-9009
Gene Expression (rlu/mg)
2500000
2000000
1500000
1000000
500000
0
1 3 1 3 1 3 1 3 1 3 1 3 1 3
Sample
Naked
Naked + PEG 5
Naked + PEG 10
CK30 PEG 5
CK30 PEG 10
C45 PEG 5
C45 PEG 10

Fig. 2

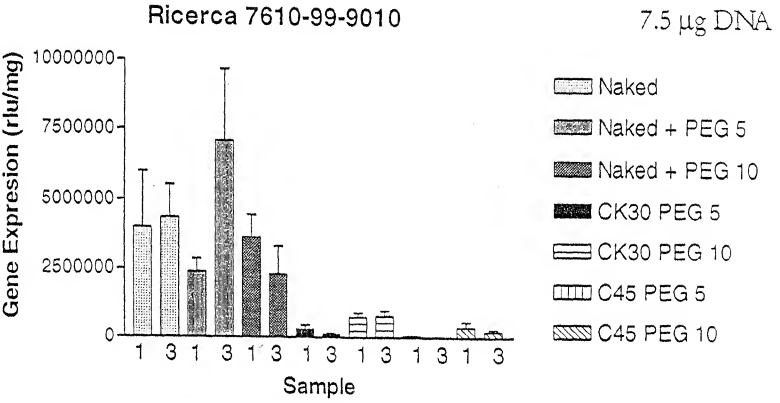

Ricerca 7610-99-9010
7.5 µg DNA
Gene Expresion (rlu/mg)
10000000
7500000
5000000
2500000
0
1 3 1 3 1 3 1 3 1 3 1 3 1 3
Sample
Naked
Naked + PEG 5
Naked + PEG 10
CK30 PEG 5
CK30 PEG 10
C45 PEG 5
C45 PEG 10

Fig. 3

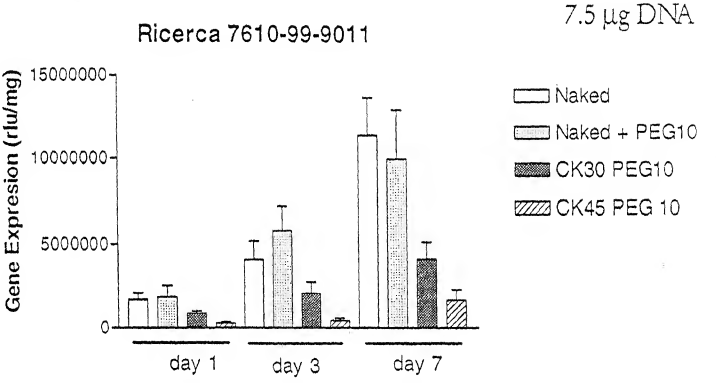

7.5 µg DNA
Ricerca 7610-99-9011
Gene Expresion (rlu/mg)
15000000
10000000
5000000
0
day 1
day 3
day 7
Naked
Naked + PEG10
CK30 PEG10
CK45 PEG 10

Fig. 4

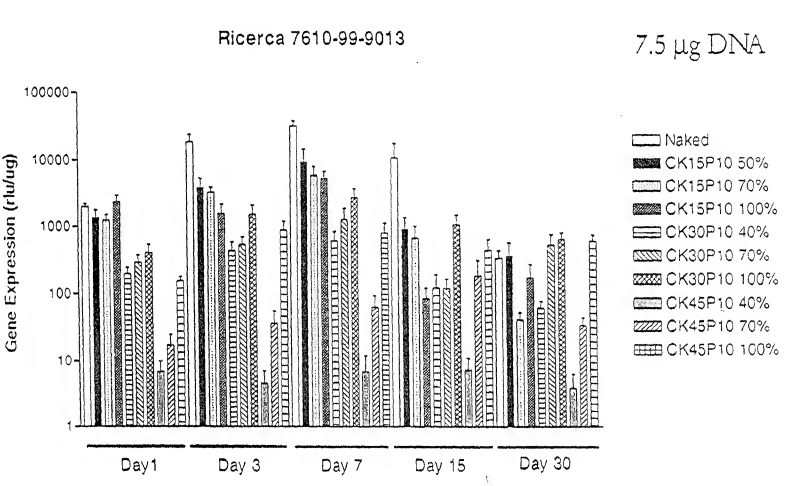

Ricerca 7610-99-9013
7.5 µg DNA
Gene Expression (rlu/ug)
100000
10000
1000
100
10
1
Day1
Day 3
Day 7
Day 15
Day 30
Naked
CK15P10 50%
CK15P10 70%
CK15P10 100%
CK30P10 40%
CK30P10 70%
CK30P10 100%
CK45P10 40%
CK45P10 70%
CK45P10 100%

Fig. 5

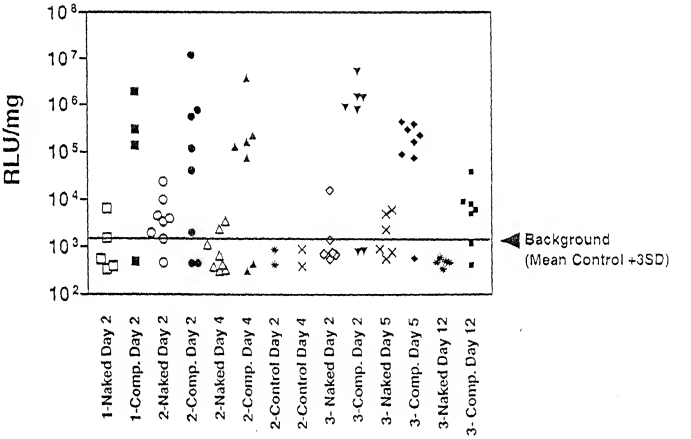

RLU/mg
10^8
10^7
10^6
10^5
10^4
10^3
10^2
Background
(Mean Control +3SD)
1-Naked Day 2
1-Comp. Day 2
2-Naked Day 2
2-Comp. Day 2
2-Naked Day 4
2-Comp. Day 4
2-Control Day 2
2-Control Day 4
3- Naked Day 2
3-Comp. Day 2
3- Naked Day 5
3- Comp. Day 5
3-Naked Day 12
3- Comp. Day 12

Fig. 6

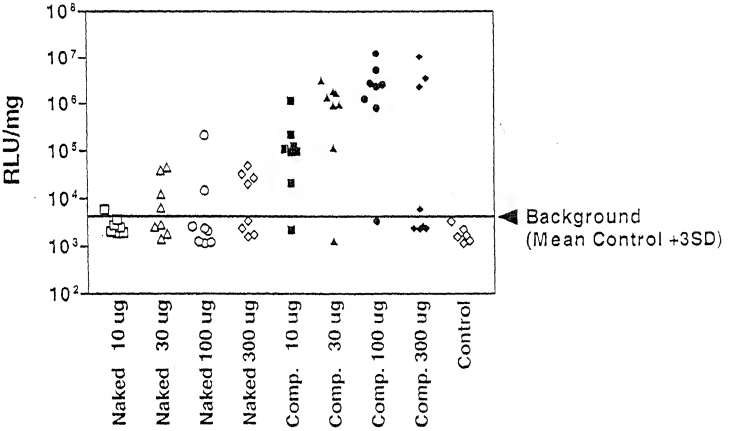

RLU/mg
10^8
10^7
10^6
10^5
10^4
10^3
10^2
Background
(Mean Control +3SD)
Naked 10 ug
Naked 30 ug
Naked 100 ug
Naked 300 ug
Comp. 10 ug
Comp. 30 ug
Comp. 100 ug
Comp. 300 ug
Control

Fig. 7A

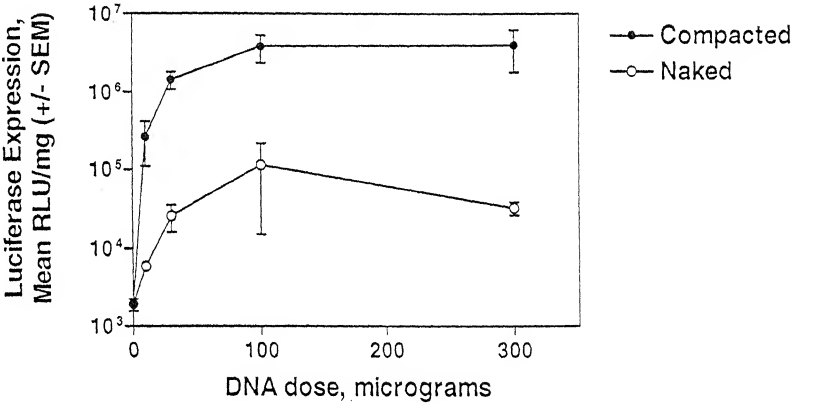

Luciferase Expression,
Mean RLU/mg (+/- SEM)
10^7
10^6
10^5
10^4
10^3
0
100
200
300
DNA dose, micrograms
Compacted
Naked

Fig. 7B

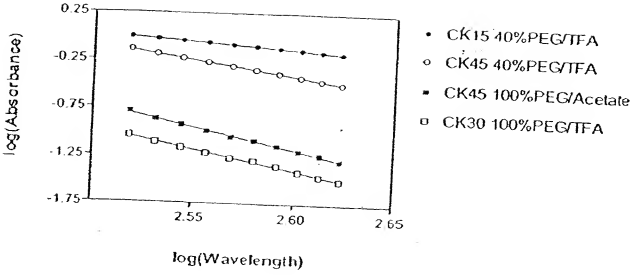
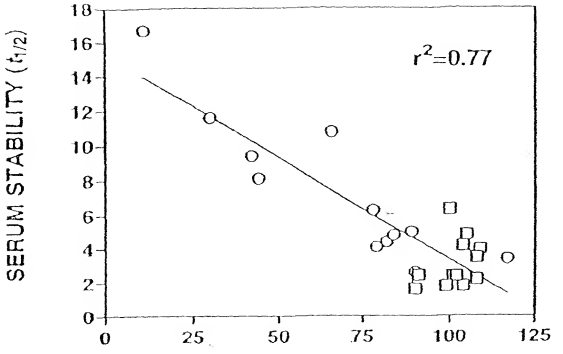

0.25
-0.25
-0.75
-1.25
-1.75
log(Absorbance)
2.55
2.60
2.65
log(Wavelength)
CK15 40%PEG/TFA
CK45 40%PEG/TFA
CK45 100%PEG/Acetate
CK30 100%PEG/TFA

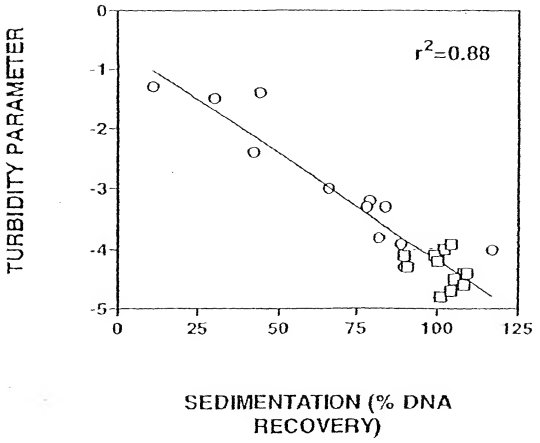
Fig. 8



o Type A Formulations

□ Type B Formulations

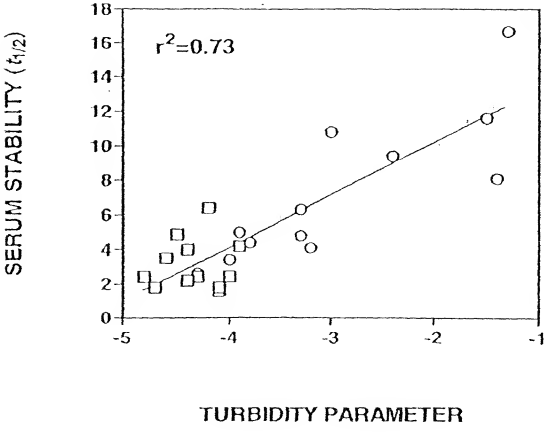
Fig. 9A



o Type A Formulations

□ Type B Formulations

Fig. 9B



○ Type A Formulations

□ Type B Formulations

Fig. 9C

PROPERTIES OF VARIOUS PLASmin™ FORMULATIONS

Formulation #	Counterion	Polylysine	PEG Content (%)	$t_{1/2}$ In Serum (h)	Turbidity Parameter	Sedimentation (%)
1	TFA	CK_{15}	40	11.6	-1.5	30
2			60	10.8	-3.0	66
3			80	9.4	-2.4	42
4			100	16.7	-1.3	11
5	TFA	CK_{30}	40	8.1	-1.4	44
6			60	4.1	-3.2	79
7			80	3.4	-4.0	117
8			100	2.6	-4.3	90
9	TFA	CK_{45}	40	6.3	-3.3	78
10			60	4.4	-3.8	82
11			80	4.8	-3.3	84
12			100	5.0	-3.9	89
13	Acetate	CK_{15}	40	2.4	-4.8	101
14			60	1.8	-4.7	104
15			80	1.6	-4.1	90
16			100	2.4	-4.0	102
17	Acetate	CK_{30}	40	3.8	-4.1	99
18			60	2.4	-4.3	93
19			80	2.2	-4.8	108
20			100	4.0	-4.4	109
21	Acetate	CK_{45}	40	6.4	-4.2	100
22			60	4.2	-3.9	104
23			80	4.9	-4.5	105
24			100	3.5	-4.6	108

Fig. 9D

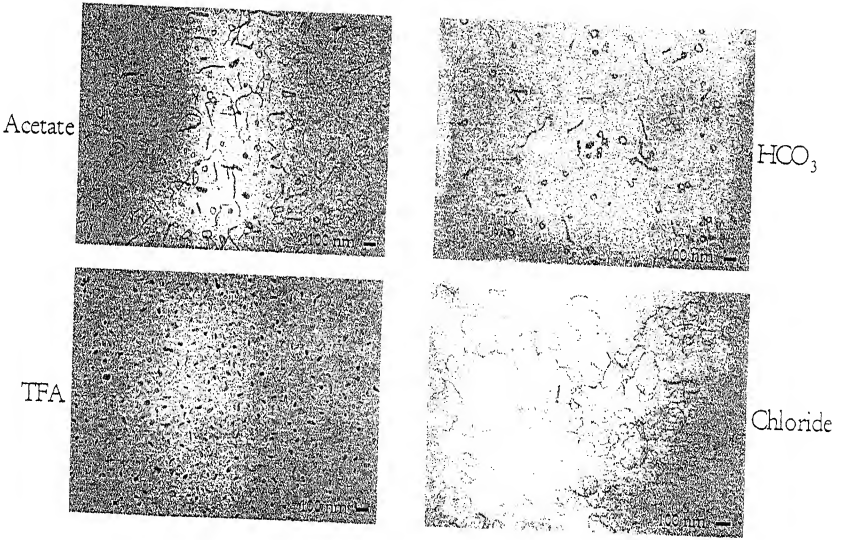


Fig. 10

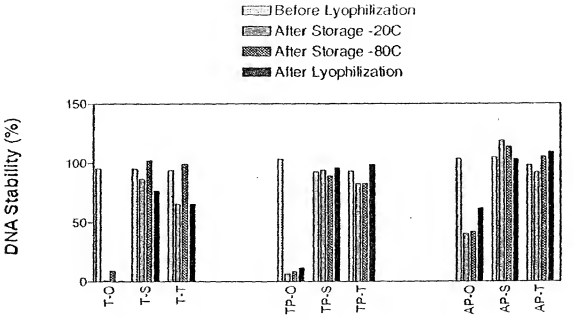
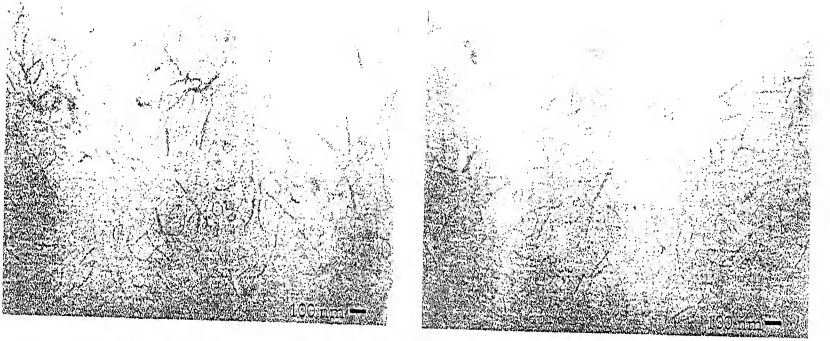

Before Lyophilization
After Storage -20C
After Storage -80C
After Lyophilization
DNA Stability (%)
150
100
50
0
T-O
T-S
T-T
TP-O
TP-S
TP-T
AP-O
AP-S
AP-T

Fig. 11

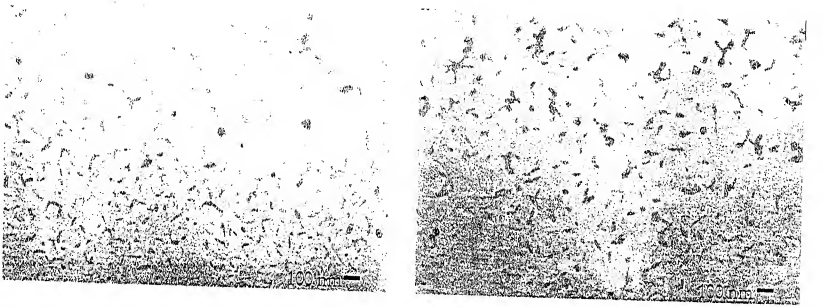
Sample	Before Lyophilization	After Lyophilization
CK30TFA		
Original		
0.5M Sucrose	-4.31	ppt
0.5 M Trehalose	-3.81	-4.10
CK30P10k - TFA	-4.70	-4.01
Original		
0.5M Sucrose	-4.51	NE-4.61
0.5 M Trehalose	-4.15	
CK30P10k - Acetate	-4.65	-4.66
Original		-3.86
0.5M Sucrose	-4.76	
0.5 M Trehalose	-4.56	-3.32
	-4.57	-4.39

Fig. 12



BEFORE AFTER

Fig. 13



BEFORE AFTER

Fig. 14

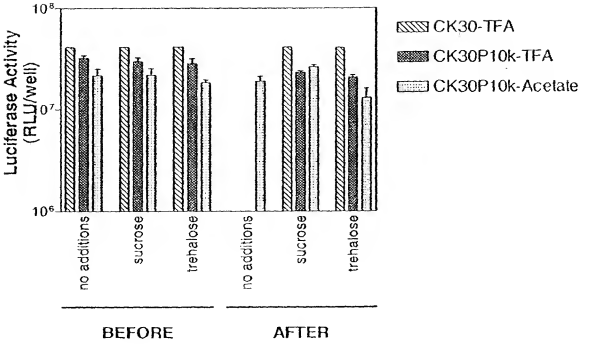

Luciferase Activity
(RLU/well)
10^8
10^7
10^6
CK30-TFA
CK30P10k-TFA
CK30P10k-Acetate
no additions
sucrose
trehalose
no additions
sucrose
trehalose
BEFORE
AFTER

Fig. 15

Polylysine	Counterion	DNA Recovery	Turbidity Parameter
CK30P10k	Acetate	100	-4.2
	Bicarbonate	98	-4.0
	Chloride	101	-5.2'
	TFA	97	-4.6
CK45P10k	Chloride	105	-4.0

Fig. 16

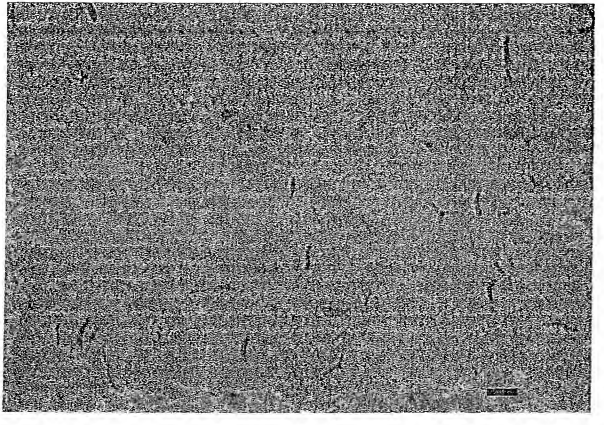


Fig. 17

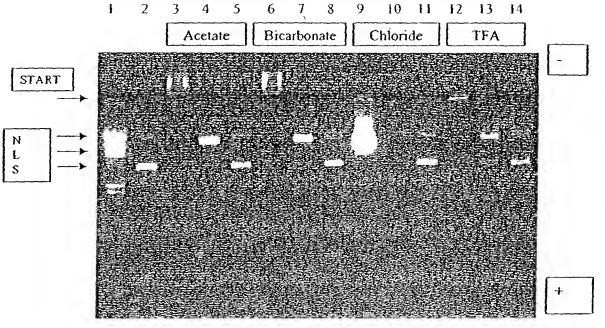

1 2 3 4 5 6 7 8 9 10 11 12 13 14
Acetate
Bicarbonate
Chloride
TFA
START
N
L
S
-
+

Fig. 18

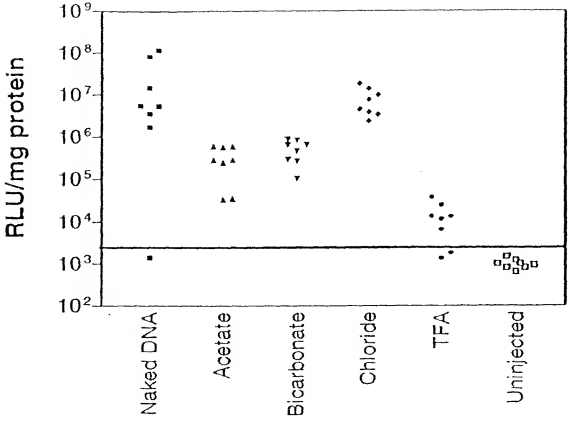

RLU/mg protein
10^9
10^8
10^7
10^6
10^5
10^4
10^3
10^2
Naked DNA
Acetate
Bicarbonate
Chloride
TFA
Uninjected

Fig. 19

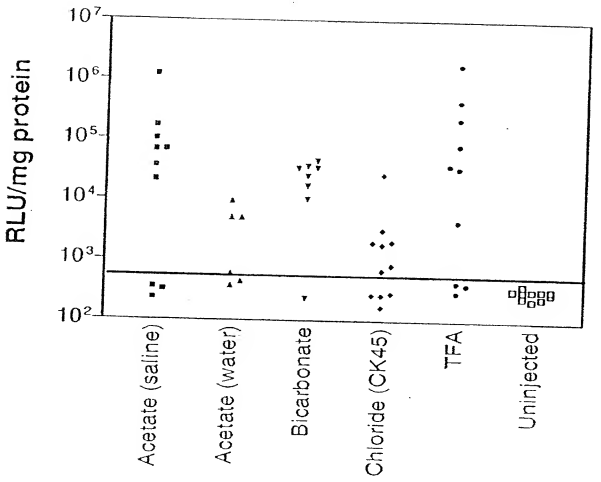

RLU/mg protein
10^7
10^6
10^5
10^4
10^3
10^2
Acetate (saline)
Acetate (water)
Bicarbonate
Chloride (CK45)
TFA
Uninjected

Fig. 20